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REVIEW

Apelin, diabetes, and obesity

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Abstract Apelin is a peptide known as the ligand of the G-protein-coupled receptor APJ. Several active apelin forms exist such as apelin-36, apelin-17, apelin-13, and the pyroglutamated form of apelin-13. Apelin and APJ are expressed in the central nervous system, particularly in the hypothalamus and in many peripheral tissues. Apelin has been shown to be involved in the regulation of cardiovascular and fluid homeostasis, food intake, cell proliferation, and angiogenesis. In addition to be an ubiquitous peptide, apelin is also produced and secreted by adipocytes and thus considered as an adipokine. This has opened a new field of investigation establishing a link between apelin and metabolic disorders (obesity, type 2 diabetes, etc.) which is the focus of the present review. Several studies, but not all, have reported an increase of plasma apelin concentrations in humans and in animal models with different metabolic pathologies. Moreover, important roles for apelin both in glucose and lipid metabolism have been highlighted as well as the associated signaling pathways. Apelin appears as a beneficial adipokine with anti-obesity and anti-diabetic properties and thus as a promising therapeutic target in metabolic disorders.

Keywords Apelin · Insulin resistance · Obesity · Glucose metabolism · Lipid metabolism

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Apelin and its receptor APJ

The APJ receptor is a G-protein-coupled receptor (GPCR) identified in 1993 in humans, displaying a close sequence homology to the angiotensin II receptor type 1 and thus named AGTRL1 [1]. The APJ gene was mapped to chromosome 11 and later sublocalized to the locus 11q12. Its transcripts were first detected in many regions of the brain but APJ is expressed in a wide range of tissues [2]. Once cloned, APJ murine homologue was shown to emerge during embryogenesis, especially in the primary blood vessels and the forming heart [3]. APJ has been shown to inhibit forskolin-induced cAMP accumulation in transfected CHO cells [2, 4] to induce the phosphorylation of ERK, Akt, or p70 S6 kinase [5–7].

In 1998, Tatemoto and coworkers purified from bovine stomach extracts a protein that binds to the "orphan" APJ receptor [8]. Based on peptide sequences, they cloned the corresponding bovine and human cDNA. The gene encodes a 77-amino-acid polypeptide that includes a secretory signal sequence. The ligand of the orphan receptor APJ consisted in the C-terminal part of this polypeptide and was called "apelin," for *APJ Endogenous Ligand* [8]. The apelin gene has been localized on the X chromosome at Xq25-q26.3 by several mapping studies, including one reported by the group of O'Dowd who first discovered the APJ receptor [9].

Apelin gene is expressed in many peripheral tissues as well as in different brain areas (for review, see [10, 11]). Its product, namely preproapelin, was found to exist in tissues under a high molecular weight form consisting in a dimer owing to disulfide linkage [12, 13]. So far, three active forms of apelin, consisting of 13, 17, or 36 amino acids and the pyroglutamated apelin-13 (Pyr(1)-apelin-13) originating from a common 77-amino-acid pre-propeptide precursor,

have been described. The sequencing of the human, bovine, rat, and mouse preproapelin has shown that there is a high sequence homology among the four species and a perfect identity for the last 22 C-terminal amino acids [8, 9]. The predominant molecular forms of endogenous hypothalamic and plasma apelin in rats were found to be apelin-36, apelin-17, and apelin-13 [14, 15]. Pyr(1)-apelin-13 was found to be the predominant isoform in human cardiac tissue [16]. Due to a higher resistance to degradation, Pyr(1)-apelin-13 has been largely used to study in vivo or in vitro responses and is considered to be a physiologically relevant APJ ligand [10, 16]. The hydrophobic residues of apelin-13 play important roles in interactions with the APJ receptor [17]. Apelin-13 and apelin-36 differ in receptor binding affinity and in their ability to affect the intracellular trafficking of the apelin receptor [14, 18]. APJ, as a GPCR, could be internalized. This process involves recruitment of β-arrestins to GPCRs and leads to its desensitization [19]. Apelin-17 was found to be the most potent inducer of APJ internalization and the removal of a single amino-acid at the C-terminus abolished this process [4]. More recently, it has been shown that apelin-13-internalized receptor dissociated from β-arrestin1, rapidly recycled to the cell surface through a Rab-4 dependent process, while the apelin-36-internalized receptor remained associated with β-arrestin1 which may target the receptor for degradation in lysosome [20].

Given the broad range of physiological actions of apelin, APJ represents a new interesting target for pharmacological agent design. A mutation of the carboxyl-terminal phenylalanine in apelin-13 (F13A) revealed a loss of function since concomitant administration of F13A blocked the hypotensive effects of apelin-13, establishing F13A as a competitive antagonist for APJ [13]. Very recently, a novel APJ antagonist has been designed using a bivalent ligand approach [21]. One of the compounds, a cyclic peptide was shown to be a competitive antagonist of APJ and will open the investigation field for the development of further antagonists with good affinity and efficiency. A nonpeptidic agonist has also been identified using fluorescence resonance energy transfer (FRET) [22]. This agonist (E339-3D6) induced vasorelaxation of rat aorta and inhibited systemic vasopressin release in water-deprived mice [22]. Its potency did not differ from that of apelin-17 and thus E339-3D6 could represent a new generation of vasodilator and aquaretic agents.

Apelin bioavailability also deserves to be better defined, especially regarding the predominant isoforms circulating in plasma since numerous clinical studies have reported a very wide range of apelin plasma levels, in both healthy controls and in patients with different pathologies. Since standard immunoassays cannot specifically quantify each apelin peptide, liquid chromatography/tandem mass spectrometry has been adapted to quantify each plasma apelin fragment [23]. Surprisingly, the main circulating isoforms of apelin were not detected [23]. These discrepancies between the peptides quantification by immunoassay and those detected by mass spectrometry raises a number of questions and provide evidence that the nature and the concentrations of the circulating immunoreactive apelin isoforms is a very important task.

Apelin can also be degraded by the angiotensin-converting enzyme 2 (ACE2), a monocarboxypeptidase homologue to ACE [24]. ACE2 hydrolyses both apelin-13 and apelin-36 with high catalytic efficiency [24, 25]. Since different studies have shown that ACE2 is an essential regulator of heart function [26] and is involved in diabetes [27], it is of interest to study in parallel ACE2 regulation alongside with apelin action.

Apelin and APJ regulation during obesity and type 2 diabetes

Apelin in adipose tissue

Apelin has been detected in adipose tissue by Tatemoto et al. [28] and, later on, the work of Boucher et al. [29] has demonstrated that apelin was not only produced but also secreted by adipocytes. Apelin has been then considered as a new adipokine. In addition, there is a close relationship between apelin and insulin both in vivo and in vitro [29]. The expression of apelin in adipocytes is increased in various mouse models of obesity associated with hyperinsulinemia. During fasting and after re-feeding in mice, the pattern of apelin expression in adipocytes parallels the plasma levels of insulin. In the absence of insulin (streptozotocin-treated mice), apelin mRNAs in adipocytes are decreased. In cultured 3T3F442A adipocytes, insulin treatment results in increased expression and secretion of apelin [29].

Other factors regulate positively the expression of apelin in adipocytes [11]. TNFα, an inflammatory cytokine elevated during obesity-associated insulin resistance, increases apelin expression both in human and mouse adipocytes [30]. In the gastrointestinal tract, inflammation also increases apelin expression [31]. The role of apelin has not yet been fully elucidated but it could have anti-inflammatory properties [32]. Overexpression of peroxisome proliferator-activated receptor gamma (PPARγ) coactivator-1 α (PGC1α), a key regulator of cellular energy homeostasis in oxidative tissues, also induces apelin expression and secretion in human adipocyte [33]. Eicosapentaenoic acid (EPA), a polyunsaturated fatty acid (PUFA) from the omega-3 family, increased also basal and insulin-stimulated apelin secretion and gene expression in 3T3-L1

adipocytes [34]. Moreover, rats fed a cafeteria diet daily treated with oral administration of EPA ethyl ester had also a higher expression of apelin in adipose tissue [35]. Altogether, these data could be in line with the improvement of insulin sensitivity observed with PUFA treatment [36].

It has been shown that apelin expression increased during adipogenesis [29, 37] and more recently that blockade of the renin-angiotensin system (RAS) ameliorates apelin expression and secretion in 3T3-L1 adipocytes [37]. Thus, through increased apelin production, RAS blockers could prevent excessive lipid accumulation and the generation of reactive oxygen species (ROS) in differentiating adipocytes [37]. Inhibition of ROS production by apelin has also been shown in other cell types [38, 39].

Obesity is related to an increase of hypoxia in adipose tissue and increased expression of the transcriptor factor hypoxia-inducible factor 1a (HIF-1a) contribute to chronic inflammation during obesity [40]. Different studies have shown that hypoxia induced expression and secretion of apelin by both human and murine adipocytes [41–43]. Moreover, it has been demonstrated that induction of apelin under hypoxic conditions is mediated by direct HIF-1 binding to the apelin gene. Since apelin is involved in angiogenesis, which is essential for adipose tissue expansion, apelin has been proposed to contribute to the development of new vasculature in expanding fat depot [42, 43].

Curiously, negative modulators of apelin expression in adipocyte are not numerous, and only glucocorticoids (dexamethasone) have described to decrease apelin mRNA levels in 3T3-L1 cells [44].

APJ in adipose tissue

APJ is present in human and mouse adipose tissue, both in isolated adipocytes and in the stroma vascular fraction [45–48]. In contrast, data concerning APJ expression or regulation in adipocyte cell lines (3T3-L1 or 3T3F442A) are very few. With obesity, APJ expression, like apelin, is increased in human adipose tissue and this up-regulation could be reversed after diet-induced weight loss [45]. Interestingly, there is a close relationship between apelin and APJ expression in adipose tissue, and changes in insulin levels might be involved for both apelin and APJ regulations [45]. However, this regulation can be different according to the severity of insulin resistance. During fasting/refeeding transition, APJ expression is significantly increased in adipose tissue of HFD mice but not in highly insulin-resistant db/db mice. In a fed state, apelin and APJ expressions were increased in adipose tissue of HFD mice compared to control whereas in db/db mice, the level was similar to control mice [47]. In control and type 2 diabetic subjects undergoing an euglycemic-hyperinsulinemic clamp, there was no significant difference in the basal state in apelin and APJ mRNA levels and, after insulin infusion, APJ and apelin expressions were increased only in control subjects. The effect of insulin was completely blunted in adipose tissue of type 2 diabetic patients. Therefore, these data underline the need for specific clinical and longitudinal studies in humans to better define the regulation of this system in metabolic diseases.

Plasma concentrations of apelin in obese and diabetic patients

The first report in humans of plasma apelin concentrations was shown in obese and hyperinsulinemic subjects [29, 30] where plasma apelin levels are increased. Different groups also found increased plasma apelin levels in morbidly obese subjects [49], in patients without a severe obesity but with impaired glucose tolerance or with type 2 diabetes [50]. In morbidly obese patients with or without diabetes, apelin levels were only higher in the morbidly obese diabetic subjects [51]. However, reduced plasma apelin levels were described in obese subjects with untreated type 2 diabetes, compared to non-diabetic subjects [52, 53]. These results could be consistent with the fact that after 14 weeks of anti-diabetic treatment (rosiglitazone and metformin), plasma apelin levels were increased and the glycemic profile improved [54]. In women with gestational diabetes, no significant differences in plasma apelin levels were observed compared to women with normal glucose tolerance [46]. All values obtained in the cited studies are reported in Table 1. Interestingly, in gestational diabetic lactating women, apelin was present in both colostrum and mature milk, apelin concentrations being higher in mature milk [55]. In the serum of the same patients, there was a trend to lower concentrations of apelin in women with gestational diabetes compared to lactating healthy women [55]. The role of apelin in milk and the regulation involved needs to be further investigated.

Plasma apelin concentrations were also measured in obese children. In pubertal obese children, apelin as well as adiponectin levels were lower compared to non-obese children [56]. However, when comparing plasma apelin concentration in obese girls (between 14 and 18 years old) and in girls with either anorexia nervosa or with no otherwise specified eating disorders, apelin concentrations were significantly higher in obese compared to healthy control but lower in patients with both eating disorders compared to healthy control [57]. Recently, no difference in apelin levels was found and no significant correlations between apelin and weight status, body fat, insulin

Table 1 Plasma apelin concentrations in adults obese and type 2 diabetic patients

Patients, n = number of case	Apelinemia (pg/ml)	BMI (kg/m^2)	Reference
Control, $n = 8$	170.6 ± 23.8	23.4 ± 0.6	Boucher et al. [29]
Obese, $n = 9$	250.9 ± 24.2	32.6 ± 0.4	
Control, $n = 12$	272 ± 20	20.7 ± 0.6	Castan-Laurell et al. [45]
Obese, $n = 20$	369 ± 25	32.2 ± 6.4	
Control, $n = 12$	174 ± 14	22 ± 2	Heinonen et al. [49, 60]
Morbide obese, $n = 25$	736 ± 50	48 ± 1	
Control, $n = 12$	1120 ± 510	24.9 ± 3.1	Soriguer et al. [51]
Morbide obese, $n = 15$	1070 ± 103	51.5 ± 7.3	
Obese and diabetic, $n = 16$	1870 ± 122	53.9 ± 7.5	
Control, $n = 36$	389 ± 17	23.0 ± 3.2	Li et al. [50]
Glucose intolerant, $n = 26$	459 ± 32	23.9 ± 3.4	
Diabetic, $n = 30$	498 ± 35	23.5 ± 2.4	
Control, $n = 11$	101.7 ± 3.6	23.0 ± 0.4	Dray et al. [47, 66]
Diabetic, $n = 12$	127.8 ± 11.6	31.0 ± 1.3	
Control, $n = 40$	750 ± 420	28.9 ± 3.3	Erdem et al. [52]
Untreated diabetic, $n = 40$	440 ± 380	29.9 ± 3.1	
Control, $n = 101$	1656.5[a]	22.9[a]	Telejko et al. [46]
Gestational diabetes, $n = 101$	1555.6[a]	23.8[a]	

[a] Median value

resistance, and cardiovascular risk factors associated with obesity between 80 obese and 40 lean children [58]. In children with type 1 diabetes, plasma apelin levels were increased compared to non-diabetic subjects [59] suggesting that the lack of insulin in this situation has no impact on apelin levels.

Changes in apelin levels after weight loss or bariatric surgery in obese individuals were also investigated. Diet-induced weight loss decreases apelin levels in moderate obese women [45] but not significantly in patients with the metabolic syndrome [60] or in obese children [58]. Bariatric surgery resulted in a significant decrease in apelin levels only in morbidly obese patients exhibiting impaired fasting glucose or type 2 diabetes before surgery [51].

All together, these studies underline that obesity, per se, is probably not the main determinant of increased plasma apelin concentrations since circulating apelin levels are not necessary significantly correlated to the body mass index (BMI) in all the published studies [46, 51, 55, 58]. However, plasma apelin or changes in plasma apelin concentrations were found to correlate significantly with serum triglycerides, glucose [51], TNFα [60], HOMA-IR, (Homeostasis Assessment Model of Insulin Resistance) [45, 50, 61], and HbA1c [47].

Finally, a polymorphism study performed in China on 3,700 subjects (1,892 patients with type II vs. 1,800 controls) described a strong association between a variant of the gene for apelin and plasma levels of fasting glucose in the Han population [62].

Effect of apelin on insulin secretion

The first evidence of an involvement of apelin on insulin secretion came from the study of Sorhede Winzell et al. showing that apelin inhibits insulin secretion stimulated by glucose in vivo in mice and in vitro in isolated islets of Langerhans [63]. Apelin-36 was used in this study and had no glucose-lowering effect by itself. More recently, apelin-13 was also shown to inhibit insulin secretion stimulated by high glucose concentrations (10 mM) or potentiated by GLP-1 in INS-1 cells [64]. The intracellular pathway activated by apelin involves a decrease of cAMP (or GLP-1-stimulated cAMP production) in the β-cells by a PI3-kinase-dependent activation of phosphodiesterase 3B rather than the inhibition of adenylyl cyclase. Interestingly, the dose-effect of apelin was biphasic, and maximal stimulation was reached at 100 nM [64, 65]. It could be speculated that in hyperinsulinemic obese subjects, the high levels of plasma apelin failed to decrease insulinemia.

Peripheral effects of apelin on energy metabolism

Glucose metabolism

Apelin effects in standard mice

Intravenous apelin administration at low concentration (200 pmol/kg) decreased blood glucose in mice and

improved glucose tolerance [66]. Furthermore, during an hyperinsulinemic-euglycemic clamp, when the hepatic glucose production is totally inhibited, apelin increases glucose utilization throughout the entire organism mainly due to a rise in glucose uptake by skeletal muscles and adipose tissues. In isolated skeletal muscle (soleus), apelin stimulates glucose transport and its effect is additive to that of insulin [66]. The associated signaling pathway involved was shown to be dependent of AMP-activated protein kinase (AMPK) and of endothelium NO synthase (eNOS) activation. AMPK is a key enzyme of energy metabolism activated during ATP depletion in cells. It is involved in various metabolic processes stimulating the production of energy such as glucose transport [67]. We demonstrated by both in vivo and in vitro experiments that AMPK was phosphorylated by apelin in soleus muscle and involved in apelin-stimulated glucose transport [66]. More recently, in cultured C2C12 myotubes, apelin-induced glucose uptake was also shown to be dependent of AMPK activation [68]. In addition, like insulin, apelin phosphorylates Akt and its activation is necessary for glucose transport both ex vivo in soleus muscle and in C2C12 myotubes. Moreover, the activation of Akt is AMPK dependent [66, 68].

Apelin was also shown to stimulate glucose transport in an AMPK-dependent manner in human adipose tissue [48]. Moreover, in insulin-resistant 3T3-L1 adipocytes (due to TNFa treatment for 24 h), insulin-stimulated glucose uptake was reduced by 47%, whereas apelin treatment resulted in an increased glucose uptake through the PI3K/Akt pathway and improved insulin-stimulated glucose uptake [69].

Apelin effects in obese and insulin-resistant mice

Mice fed a high-fat diet (HFD) for several weeks become obese, hyperinsulinemic, and insulin resistant. Glucose tolerance is significantly improved in these mice receiving i.v. apelin bolus before oral glucose tolerance test (OGTT). In addition, the loss of insulin sensitivity observed during a euglycemic-hyperinsulinemic clamp in insulin-resistant mice was improved with an apelin perfusion during the clamp [66]. Thus, apelin acute treatment is still efficient in obese insulin-resistant mice and improves the altered glucose metabolism especially by increasing glucose uptake in skeletal muscle. Very recently, chronic apelin treatment in insulin-resistant mice was shown to improve insulin sensitivity [68]. The role of apelin in glucose homeostasis was also confirmed by the phenotype of apelin null mice that are hyperinsulinemic and insulin resistant. The loss of insulin sensitivity in apelin −/− mice was exacerbated by a high fat/high sucrose diet [68].

Thus, apelin is positively involved in carbohydrate metabolism and displays beneficial properties such as glucose-lowering effects (Fig. 1). Although plasma apelin levels are elevated in obese insulin-resistant mice, exogenous apelin is still efficient and thus apelin resistance

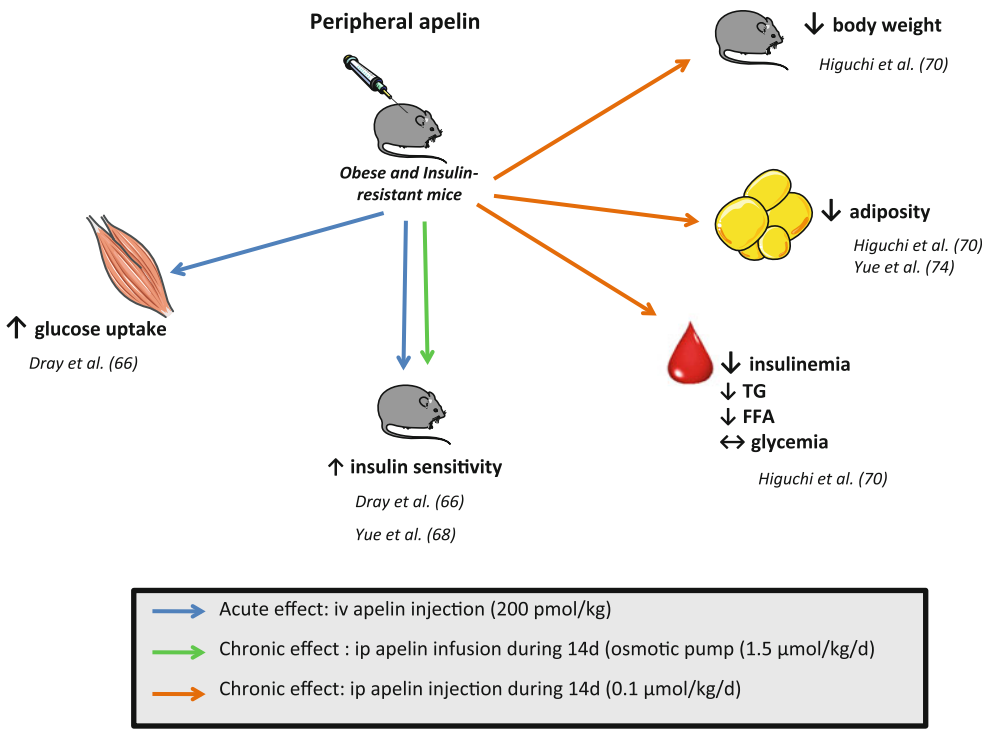

Fig. 1 Metabolic effects of peripheral apelin after acute or chronic treatment in obese and insulin-resistant mice. Apelin has been administered either by intravenous injection (i.v.) during acute treatment or by daily intraperitoneal injection (i.p.) or via osmotic pump during chronic treatment. *TG* triglycerides, *FFA* fatty acids

unexpected. We hypothesized that the increased levels of apelin might constitute a compensatory mechanism to delay the onset of insulin resistance.

Lipid metabolism

Apelin effects in standard rodents

The first study that reported a role of apelin on lipid metabolism was related to chronic peripheral administration of apelin (during 2 weeks) in standard mice [70]. Daily i.p. apelin injection was shown to decrease the triglycerides content in adipose tissue and the weight of different fat depots and in chow-fed mice [70]. A decreased adiposity was also found in obese mice. Plasma triglycerides were also decreased in both normal and obese apelin-treated mice. The treatment did not affect average food intake but increase rectal temperature and O_2 consumption. An increased expression of mitochondrial uncoupling protein 1 (UCP1) was observed in brown adipose tissue (BAT) (Higuchi 2007). All together, the authors suggest that apelin increases energy expenditure through UCP1 activation. In addition to changes in BAT UCP1, apelin treatment also increased UCP3 expression in skeletal muscle but no metabolic effects were measured [70]. Since increase of UCP3 content in muscle could result in an increase of mitochondrial biogenesis, a recent study was conducted in rat in order to know whether chronic apelin treatment would lead to an increase in mitochondrial enzyme activity and protein content in skeletal muscle [71]. Accordingly, enzyme activities of β-HAD (involved in the mitochondrial oxidative capacities), citrate synthase (involved in the citric acid cycle), and cytochrome C oxidase (COX) (involved in the respiratory chain) were increased in response to apelin treatment in rat triceps. Surprisingly, the increase of mitochondrial markers was independent of increased PGC-1α expression, identified as a key player in the regulation of mitochondrial biogenesis. However, PGC-1β was up-regulated in triceps muscle. The role of PGC-1β in muscle is less clear than the one of PGC-1α but very recently, Wright et al. provide evidence, by overexpressing PGC-1β in muscle of HFD rats, that it has a protective effect against lipid-induced insulin resistance [72]. Thus, apelin treatment during insulin resistance through PGC-1β activation could improve mitochondrial oxidative capacities.

Apelin effects in obese and insulin-resistant mice

Chronic apelin treatment decreased adiposity and similar results were obtained in mice over-expressing apelin (apelin-transgenic (apelin-Tg) mice) fed a HFD. Apelin-Tg mice exhibited a resistance against diet-induced obesity, increased oxygen consumption and body temperature without modification of food intake [73]. Interestingly, diet-induced obesity resistance of apelin-Tg mice was correlated with vascular formation in skeletal muscle due to an increase of angiopoietin-1 and its receptor Tie 2 [73]. These mice present also increased mitochondrial biogenesis in skeletal muscle but by a PGC-1α-independent pathway. The involvement of PGC-1β has not been studied.

In agreement with the phenotype of apelin-Tg mice, apelin −/− (APKO mice) mice had increased abdominal adiposity and increased circulating FFA levels [74]. After reintroduction of exogenous apelin (apelin infusion during 2 weeks) in APKO mice, adiposity and fatty acids but also glycerol levels were decreased in apelin-treated APKO mice, suggesting a role of apelin in lipolysis regulation. In both isolated adipocytes and 3T3-L1 differentiated adipocytes, apelin was shown to inhibit isoproterenol- (β-adrenergic agonist) induced lipolysis [74] through a pathway involving Gq, Gi, and AMPK. However, in human adipose tissue explants or isolated adipocytes, apelin had no effect on basal or isoproterenol-stimulated lipolysis even though apelin was shown to activate AMPK in human adipose tissue [48].

A question to be addressed is the outcome of lipids. Modification in the lipid metabolism conducting to excess fatty acid accumulation in non-adipose tissues is a hallmark of metabolic diseases. Our group is presently investigating the oxidative capacities of skeletal muscles in response to apelin treatment since apelin activates AMPK in muscle [66]. Interestingly, chronic apelin treatment increases fatty acid β-oxidation in soleus muscle especially in HFD fed mice [75]. These data are in agreement with the fact that apelin treatment improves insulin sensitivity.

Central effects of apelin on energy metabolism

Both apelin and its receptor APJ have been detected throughout the central nervous system particularly in the hypothalamus. Apelin mRNAs are present in different nuclei including the paraventricular, arcuate, and supraoptic nuclei that are involved in the control of behavioral, endocrine processes, and energy homeostasis [76]. Apelin-positive nerve fibers in the hypothalamus imply the existence of apelinergic neurons and thus a dual action of apelin as a circulating peptide and a neurotransmitter. So far, it is not known whether peripheral plasma apelin can reach the hypothalamus and could modulate apelin levels in the hypothalamus. Higuchi et al. mentioned that apelin concentration in the hypothalamus is increased after apelin i.p. injection [70].

The effects of acute intracerebroventricular (i.c.v.) apelin administration on food intake and energy expenditure were mainly studied in rats and the results are

contradictory. Apelin i.c.v. was shown to decrease food intake in fed and fasted rats [77] and during nocturnally administration of apelin whereas during day-time apelin stimulates feeding [78]. Furthermore, no significant effect was reported on the accumulated 24 h food intake in rat [79]. More recently, Clark et al. showed that i.c.v. apelin injection decreased food and water intake and respiratory exchange ratio in control rats, but had no effect in high-fat fed rats [80]. Moreover, apelin induced a down-regulation of central APJ receptor only in HF-fed rats, suggesting that a decreased central response to apelin could induce obesity [80]. In addition, an increase of core body temperature and locomotor activity was also observed after central apelin injection [81]. In mice, only one study measured the same effects as well as food intake after a chronic 10 day i.c.v. infusion of apelin-13 (1 μg/day) into the third ventricle. Apelin treatment increased food intake (on day 3 to 7), locomotor activity especially during the nocturnal period when feeding occurs and body temperature only during the period of activity [82]. Moreover, these mice had increased adiposity [82].

The role of central apelin on glucose metabolism has been recently studied in our group. Acute i.c.v. apelin has differential effect depending of the injected dose and the nutritional status [83]. Acute low-dose of i.c.v. apelin injection decreased peripheral fed glycemia, increased glucose and insulin tolerance in mice via a NO signaling pathway. All these beneficial actions of i.c.v. apelin on glucose homeostasis were blunted in HFD obese/diabetic mice. As the opposite, acute high-dose of i.c.v. apelin injection provoked fasted hyperglycemia/hyperinsulinemia and decreased insulin sensitivity in normal mice. These effects are summarized in Fig. 2. Moreover, acute high-dose of i.c.v. apelin injection in HFD mice increased fasted hyperglycemia. Thus, elevated levels of central apelin might impair glucose homeostasis since obese/diabetic mice, which are sensitive to peripheral apelin [66], have increased plasma apelin levels [83]. Both the finding of abolished circadian apelin regulation in HFD-treated mice and that of chronic apelin treatment in normal mice triggering insulin intolerance are consistent with this hypothesis [83]. Other parameters explaining this differential action of apelin is the fact that hypothalamic nuclei (ventromedian hypothalamus, dorsomedian hypothalamus and arcuate nucleus) had different modes of activation in term of c-Fos expression in response to i.c.v. apelin. Similar to the control of peripheral glycemia/insulinemia, hypothalamic neuronal activation by apelin varied according to the injected dose (low vs. high) of apelin and the nutritional status (fasted vs. fed) [83]. Actually, we hypothesized that a rise in hypothalamic apelin levels could be involved in the transition from normal to diabetic status.

Conclusion

Apelin has pleiotropic effects on numerous organs and tissues but recent experimental investigations have enhanced the role of apelin on whole body metabolism (Figs. 1, 2). Several studies have reported a protective

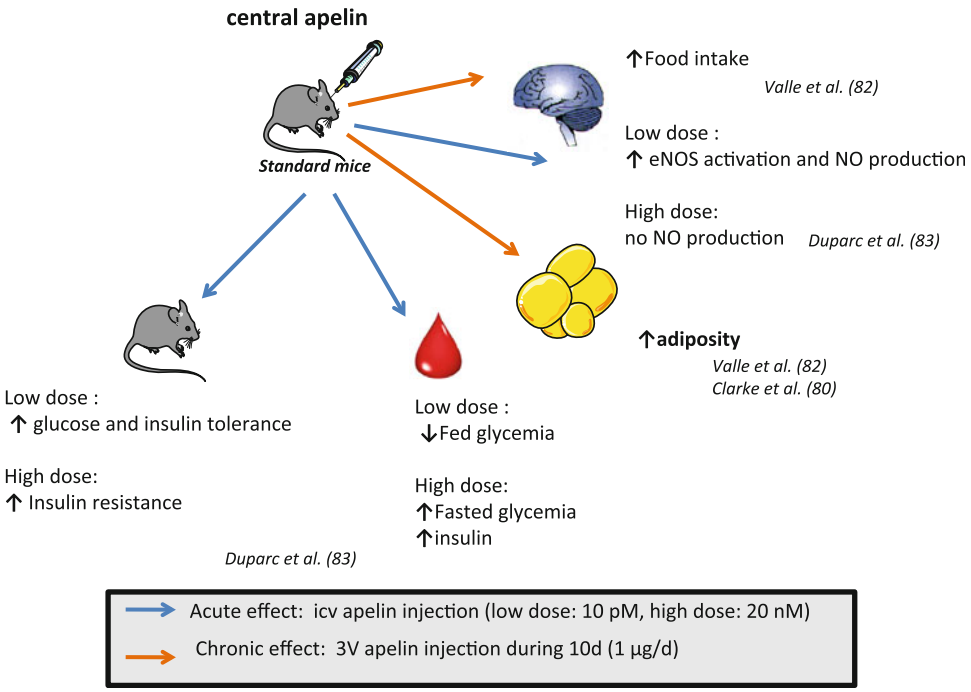

Fig. 2 Metabolic effects of central apelin after acute or chronic injection in standard mice. Apelin has been administered by intracerebroventricular injection (i.c.v.) during acute treatment or by daily injection in the third ventricule (3v) during chronic treatment

action of apelin in obesity-associated diseases [84] notably, a cardio-protective effect and now, apelin, like adiponectin, can be considered as an insulin-sensitizing agent. The insulin mimetic effects of apelin on glucose metabolism are very encouraging and of major interest in the context of a therapeutic approach in type 2 diabetic patients. Studies in humans will be essential to confirm the role of apelin on carbohydrate metabolism. In addition, accumulating evidence suggests that apelin is also an important regulator on lipid metabolism by reducing fat mass and promoting fuel consumption, which is also consistent with an improvement of insulin sensitivity. Those findings strengthen the beneficial effects of apelin in obesity-associated diseases and hoist it on the list of potent therapeutic target.

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Conflict of interest The authors declare that they have no conflict of interest.

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